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From the Editor - Happy New Year! We have been doing the *ag animal health* newsletter for over eight years now. How time flies. Across the years we have devoted a lot of space to food safety in our livestock species, specifically articles on antibiotic residues and antimicrobial resistance. This issue will deal once more with antimicrobial resistance as we (Extension specialists and Educators) work this year to get producers and veterinarians in Washington ready for the rules associated with the Veterinary Feed Directive.
DA Moore

The ag animal health newsletter is devoted to the transfer of current, relevant information to food animal owners and veterinarians.

Congratulations are in Order!



Dr. Tim Baszler, former Director of WADDL, our diagnostic laboratory, is now the Executive Director of WADDL and is also working in the Paul G. Allen School for Global Animal Health on disease surveillance.



Dr. Tom Besser, Professor, Veterinary Microbiology and Pathology, has been named to an endowed chair position. He will serve as the Rocky Crate D.V.M. and Wild Sheep Foundation Endowed Chair in Wild Sheep Disease Research

The Veterinary Feed Directive – Not just for beef!

By Dale A. Moore, Extension Veterinarian

Although the Veterinary Feed Directive (VFD) will not be official until December of this year (2016), now is the time to start thinking and acting on in-feed and in-water uses of antimicrobials. Veterinarians and food animal owners should meet to go over the products used on the farm and develop a plan on overall use of antimicrobials knowing that all medically important antibiotic use in feed and water will either require a VFD or an RX (for water soluble medications).

No more use of antibiotics for growth promotion. That means pigs, calves, lambs and chickens... All food animals. The antibiotics are to be used for treatment when animals are diagnosed with an illness, prevention of illness when exposure is likely, or control of the spread of disease. New drug labels will include "Caution: Federal law restricts medicated feed containing this veterinary feed directive (VFD) drug to use by or on the order of a licensed veterinarian." There is no extra-label use of VFD drugs allowed.

The VFD drugs are not prescription drugs. However, the first step is similar, that is to establish a valid VCPR - Veterinary Client Patient Relationship -- with a veterinarian licensed to practice in the state. For a valid VCPR, the veterinarian knows the clients, the animals and the conditions for which the drug is being used. There needs to be an initial site visit (not by telephone or email) to establish the VCPR. The veterinarian must assume responsibility for making clinical judgments regarding the health of the animal(s) and need for medical treatment, and the client must follow the instructions of the veterinarian. The veterinarian must be readily available for follow-up evaluation or has arranged for emergency coverage and continuing care and treatment.

For the VFD, the veterinarian can prepare a written or electronic directive for in-feed use of an antibiotic for treatment, prevention or control of disease. The veterinarian submits the VFD order to the feed distributor. The directive must include the expiration date -- the time by which the drug must be fed (maximum 6 months). If the feed is not fed before the expiration date arrives, a new VFD must be written. The directive must include the duration of use, the amount of time the VFD feed can be fed - it is not for continuous use. The VFD includes a lot of additional information that is specific to the veterinarian, location of, condition and animals being treated (See the FDA website below for all the required information). The veterinarian, feed distributor and client all maintain the record for two years.

What drugs are included? Those drugs that are deemed medically important to humans and animals will require a VFD if fed. They include: penicillins, cephalosporins, macrolides, quinolones, sulfas, glycopeptides, fluoroquinolones, tetracyclines and some others, such as neomycin. The ionophores are NOT included because they are not medically important.

For dairy cattle, some considerations are to evaluate feeding of medicated milk replacer and starter grain. These uses will require a VFD. For older heifers, use of tetracycline products fed for pneumonia treatment, prevention and control will require a VFD and farmers will be required to follow product labels for dose and duration. Tetracycline and other drugs used for hoof treatments will require a prescription.

What about 4H projects? Lambs, calves and pigs are raised by 4H youth and most often enter the food supply. These project animals are no different from other animals raised for food. A VFD is required for all in-feed antibiotics that are considered medically important.

All the FDA information on the Veterinary Feed Directive can be found at the FDS website: <http://www.fda.gov/AnimalVeterinary/DevelopmentApprovalProcess/ucm071807.htm>
For Additional Resources on the Veterinary Feed Directive for veterinarians, go to <http://www.aabp.org/> for the member site on VFD Resources.



Porcine Epidemic Diarrhea: It's in Oregon!

January 2016 by Sarah Smith, WSU Extension, Brad R. LeaMaster, State Veterinarian, Oregon Department of Agriculture, Dr. Charles Estill, Extension Veterinarian, Oregon State University, Dr. Jerry Heidel, Director, OSU Diagnostic Laboratory, Gene J. Pirelli, Extension Swine Specialist, Oregon State University, Dr. Joe Baker, State Veterinarian, WA Department of Agriculture

The Oregon State Veterinarian, Dr. Brad LeaMaster D.V.M., has confirmed the presence of Porcine Epidemic Diarrhea virus (PEDv) on an Oregon farm in Clackamas County. The infected premise has been quarantined and Oregon Department of Agriculture is conducting an epidemiology investigation to the sourced farm. Washington State Veterinarian, Dr. Joe Baker D.V.M., has been in contact with Dr. LeaMaster and is monitoring the situation. At this time, Dr. Baker indicated that there is no cause for alarm for pig farms in Washington.

PEDv poses no danger to other livestock, human or the safety of pork products. But it has devastating effects on suckling pigs, causing significant losses (up to 100% mortality) in pigs less than two weeks of age. Older pigs and sows will recover from the disease with some impact on performance.

PEDv was first confirmed in the U.S. in the spring of 2013 in the Midwest. During the winter of 2013-2014 it was estimated that 7-8 million piglets died national resulting in significant economic losses to farms. The national PEDv outbreak has subsided, however prior to this

Oregon break the other closest states with confirmed PEDv infection was Montana and southern Idaho. PEDv is a highly transmissible disease. Most farms infected by PEDv report production losses (baby pig deaths) for approximately 5 weeks. In addition to the significant economic loss, this disease took a heavy emotional toll on producers because of the significant death loss and lack of being able to prevent the disease even with stepped up biosecurity measures.

Vehicles with manure contamination are the most likely means of spreading the virus. Biosecurity measures in place should include limiting movement of people, equipment, and vehicles on and off the farm; thoroughly disinfect items leaving and entering the farm; establish designated routes and parking areas for vehicles; isolate new animals to the farm; and dedicate footwear and outerwear for people having contact with the animals. The National Pork Board has an excellent set of resources concerning PEDv available at <http://www.pork.org/pork-checkoff-research/pedv/pedv-resources/> . Resources cover PEDv basic and diagnosis, on-farm strategies, transportation strategies, manure strategies, and show pig strategies. In addition, Washington State Veterinarian Dr. Joe Baker will be discussing swine health issues in Washington at the Washington State Swine Information Day on February 5 in Moses Lake; more information is available at <http://www.animalag.wsu.edu/swine/WASwineInfoDayRegistration2016.pdf> .

There are two commercially available vaccines. The Harris Vaccine Company markets iPED (virus subunit) under a conditional license from the USDA. Zoetis also has a PEDv vaccine (killed virus). These vaccines are to be used in pregnant gilts and sows, NOT baby pigs. The concept involves stimulating the maternal immunity that is passed to the newborn pigs when they suckle colostrum. This may provide protection for the piglets for a few weeks. The initial vaccination should be at 5 and 2 weeks pre-farrowing then 2 weeks pre-farrowing for subsequent litters. It appears the vaccine is most effective for sows that have been previously exposed to PEDv rather than naïve, non-exposed females. Vaccination should only be viewed as an aid in prevention of disease and not a substitute for excellent biosecurity.

Pig farmers are encouraged to maintain strict biosecurity measures on their farm and when purchasing pigs. Owners should contact their veterinarian for assistance with diagnosis, vaccination, disinfection, and other PEDv mitigation details. In addition, any swine shipments entering Washington must be accompanied by a valid Certificate of Veterinary Inspection (CVI or aka health certificate) declaring that the pigs have not been exposed to PEDv and disease free. The certificate must declare the pigs did not come from anywhere known to be infected with PEDv. The certificate must be signed by both a licensed veterinarian and the animal owner. This Certificate of Veterinary Inspection (CVI) is required for all pigs entering Washington not going directly to slaughter, so this included show pigs, feeder pigs, breeding pigs, pet pigs, etc. If you have questions about importation of pigs into Washington please contact the Washington State Department of Agriculture Animal Service Division at (360) 902-1881.

[For a summary of PEDv, see the Fall 2015 issue of *ag animal health*:
<http://vetextension.wsu.edu/newsletters/> .]

Avian Influenza – Starting Already in the US

Highly Pathogenic Avian Influenza (HPAI) was identified in a turkey flock in Dubois County, Indiana. This is a different strain of HPAI (H7N8) than those that caused the 2015 outbreak. Surveillance sampling in the 10 km zone found 9 infected flocks. All of the affected flocks so far have been turkey operations. All but one of those additional affected flocks were infected with low-path H7N8, never a problem before. This particular virus is a low-path virus in waterfowl, but can mutate quickly to a highly-pathogenic form in poultry, requiring a change in a single gene. All affected flocks are being depopulated, and a high-risk layer operation adjacent to the index farm was also being depopulated.

No cases of HPAI H7N8 virus infection have been reported in humans at this time, and no human infections associated with avian influenza A viruses of this particular subtype (i.e., H7N8) have ever been reported. See the USDA APHIS website for more information: <http://tinyurl.com/jhk8g6v>. (Original information provided by Dr. Joe Baker, Washington State Veterinarian.)

WSU Ag Animal Health Research Abstracts

1) Sheng H, Shringi S, Baker KN, Minnich SA, Hovde CJ, Besser TE. Standardized E. coli O157:H7 exposure studies in cattle: Evidence that bovine factors do not drive increased summertime colonization. *Appl Environ Microbiol.* 2015 Nov 25. pii: AEM.02839-15. [Epub ahead of print]

Increased summertime prevalence of cattle carriage of enterohemorrhagic Shiga toxin-producing *Escherichia coli* O157:H7 (STEC O157) is associated with increased summertime incidence of human infection. The mechanism driving the seasonality of STEC O157 carriage among cattle is unknown. We conducted experimental challenge trials to distinguish whether factors extrinsic or intrinsic to cattle underlie the seasonality of STEC O157 colonization. Holstein steers (N = 20) exposed to ambient environmental conditions were challenged with a standardized pool of STEC O157 strains four times at 6-month intervals. Density and duration of recto-anal junction mucosa (RAJ) colonization with STEC O157 was compared by season (winter vs summer), dose (109 CFU vs 107 CFU), and route of challenge (oral vs rectal). Following summer challenges, RAJ STEC O157 colonization density was significantly lower (P = 0.016) and duration was shorter (P = 0.052) compared to winter challenges, a seasonal pattern opposite to that observed naturally. Colonization was unaffected by challenge route, indicating that passage through the gastrointestinal microbiome did not significantly affect the infectious dose to the RAJ. A 2-log reduction of the challenge doses in the second year trials was accompanied by similarly reduced RAJ colonization in both seasons (P < 0.001). These results refute the hypothesis that cattle are predisposed to STEC O157 colonization during summer months, either due to intrinsic factors or indirectly due to gastrointestinal tract microbiome effects. Instead, the data support the hypothesis that increased summertime STEC O157 colonization results from increased seasonal oral exposure to this pathogen.

2) Lui J, Zhao Z, Subbiah M, Call DR. Soil-borne reservoirs of antibiotic-resistant bacteria are established following therapeutic treatment of dairy calves. *Environ Microbiol.* 2015 Oct 21. doi: 10.1111/1462-2920.13097. [Epub ahead of print]

We determined if antibiotics residues that are excreted from treated animals can contribute to persistence of resistant bacteria in agricultural environments. Administration of ceftiofur, a third-generation cephalosporin, resulted in a ~3 log increase in ceftiofur-resistant *E. coli* found in the feces and pen soils by day 10 ($P=0.005$). This resistant population quickly subsided in feces, but was sustained in the pen soil (~4.5 log bacteria/g) throughout the trial (1 month). Florfenicol treatment resulted in a similar pattern although the loss of florfenicol-resistant *E. coli* was slower for feces and remained stable at ~6 log bacteria/g in the soil. Calves were treated in pens where eGFP-labeled *E. coli* were present in the bedding (~2 log/g) resulting in amplification of the eGFP *E. coli* population ~2.1 log more than eGFP *E. coli* populations in pens with untreated calves (day four; $P<0.005$). Excreted residues accounted for >10-fold greater contribution to the bedding reservoir compared with shedding of resistant bacteria in feces. **Treatment with therapeutic doses of ceftiofur or florfenicol resulted in 2-3 log/g more bacteria than the estimated ID50 (2.83 CFU/g), consistent with a soil-borne reservoir emerging after antibiotic treatment that can contribute to the long-term persistence of antibiotic resistance in animal agriculture.**

[Editor's Note: Although treating an animal may result in antimicrobial resistant bacteria that it can shed, residues that are excreted in urine or feces can affect bacteria in the soil.]

3) Kasimanickam VR, Owen K, Kasimanickam RK. Detection of genes encoding multidrug resistance and biofilm virulence factor in uterine pathogenic bacteria in postpartum dairy cows. *Theriogenology.* 2016 Jan 15;85(2):173-9.

Reckless use of antibiotics and/or development of biofilm are the rationale for the development of multidrug resistance (MDR) of pathogenic bacteria. The objective of the present study was to detect MDR genes in *Trueperella pyogenes* and to detect biofilm virulence factor (VF) genes in *Escherichia coli* isolated from the uterus of postpartum dairy cows. Uterine secretions from different parity postpartum Holstein cows ($n = 40$) were collected using cytobrush technique after a sterile procedure from cows with varying degree of uterine inflammatory conditions. The cytobrush was stored in a specimen collector, placed in a cooler with ice, and transported to the laboratory within 2 hours. The pathogens were isolated and were identified initially by their colony morphology and biochemical characteristics. To further identify and classify the single species, and to determine the presence of MDR and VF genes, the genes fragments were amplified using the respective primers by either singleplex or multiplex polymerase chain reaction protocol, and amplicons were detected by electrophoresis method. *T. pyogenes* was isolated in 17 of 40 (42.5%) cows in the study population as recognized by the 16S rRNA gene. Of the positive *T. pyogenes* samples, 8 of 17 (47.1%) were positive for integron type 1 (intl I), and none were positive for integron type 2 (intl II). Of those 8 positive for intl I, six of eight (75.0%) were positive for amplicons aadA5 and aadA24-ORF1 at 1048 and 1608 bp, respectively, associated with specific drug resistance. Presence of aadA5 indicated resistance to sulfadiazine, bacitracin, florfenicol, and ceftiofur. Presence of aadA24-ORF1 indicated resistant to sulfadiazine, bacitracin, penicillin, clindamycin, and erythromycin. *E. coli* was isolated in 18 of 40 (45.0%) cows in the study population. The genes for VF,

Agn43a, and Agn43 b, associated with biofilm production, were found in 6 of 18 (33.3%) of the positive isolates. Both T pyogenes MDR gene and E coli biofilm VF existed in more severe form of uterine diseases than subclinical endometritis. **In conclusion, 35% of T pyogenes isolates found were positive for a gene cassette associated with antibiotic resistance, and 33% of the E coli isolates contained genes for the VF associated with biofilm production.**

[Editor's Note: Metritis is a common disease of dairy cattle. The condition can sometimes be difficult to treat effectively and can result in long-term effects on fertility. Although there are a number of different kinds of bacteria that can be found in the infected uterus, Truperella and E coli are consistent findings. Finding some mechanisms for resistance to antibiotics, and their prevalence, can eventually lead to identification of the risks for them and potential mitigation of these risks.]

4) Afema JA, Mather AE, Sicho WM. Antimicrobial Resistance Profiles and Diversity in Salmonella from Humans and Cattle, 2004-2011. Zoonoses Public Health. 2015 Nov;62(7):506-17.

Analysis of long-term anti-microbial resistance (AMR) data is useful to understand source and transmission dynamics of AMR. We analysed 5124 human clinical isolates from Washington State Department of Health, 391 cattle clinical isolates from the Washington Animal Disease Diagnostic Laboratory and 1864 non-clinical isolates from foodborne disease research on dairies in the Pacific Northwest. Isolates were assigned profiles based on phenotypic resistance to 11 anti-microbials belonging to eight classes. Salmonella Typhimurium (ST), Salmonella Newport (SN) and Salmonella Montevideo (SM) were the most common serovars in both humans and cattle. Multinomial logistic regression showed ST and SN from cattle had greater probability of resistance to multiple classes of anti-microbials than ST and SN from humans ($P < 0.0001$). While these findings could be consistent with the belief that cattle are a source of resistant ST and SN for people, occurrence of profiles unique to cattle and not observed in temporally related human isolates indicates these profiles are circulating in cattle only. We used various measures to assess AMR diversity, conditional on the weighting of rare versus abundant profiles. AMR profile richness was greater in the common serovars from humans, although both source data sets were dominated by relatively few profiles. The greater profile richness in human Salmonella may be due to greater diversity of sources entering the human population compared to cattle or due to continuous evolution in the human environment. Also, AMR diversity was greater in clinical compared to non-clinical cattle Salmonella, and this could be due to anti-microbial selection pressure in diseased cattle that received treatment. The use of bootstrapping techniques showed that **although there were shared profiles between humans and cattle, the expected and observed number of profiles was different, suggesting Salmonella and associated resistance from humans and cattle may not be wholly derived from a common population.**

[Editor's Note: Pathogens like Salmonella that become resistant to antimicrobials are frustrating for human and veterinary medicine. Although antimicrobial use in animals is thought to contribute to resistance seen in people, this study demonstrated that there are some major differences in the resistance profiles of Salmonella from people compare to those from cattle.]

Continuing Education in Our Region

Veterinarians

WSU College of Veterinary Medicine Spring Conference will be April 22-23, 2016, in Pullman, WA. Eight hours of CE credit – FIVE tracks: Small animal, Equine, Food Animal, Pet Poultry and veterinary technician. For more information go to: <http://cvme.vetmed.wsu.edu>

Academy of Dairy Veterinary Consultants Spring Meeting will be April 8 and 9, 2016, Phoenix, AZ.

Producers

Country Living Expo and Cattlemen's Winterschool -- January 30, 2016, in Stanwood, WA.
<http://vetextension.wsu.edu/event/country-living-expo-cattlemens-winterschool/>

WA Swine Information Day – February 5, 2016, in Moses Lake, WA.
<http://vetextension.wsu.edu/event/wa-swine-information-day/>

Youth Swine Field Day -- March 26, 2016, at Asotin County Fairgrounds, Asotin, WA. For information, contact Janet Schmidt, Extension Whitman County at: schmidtj@wsu.edu

Cattlemen's Boot Camp – April 15 and 16, 2016, at Benton County Fairgrounds, Kennewick, WA. For Registration information, go to: <https://www.angusonline.org/event/bootcampmain.aspx>

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